REMARKS

Entry of the foregoing and further and favorable reconsideration of the subject application pursuant to and consistent with 37 C.F.R. § 1.112 is respectfully requested.

By the present amendment, claims 1, 2, 4, 5, 7-9, 11, 15, 18, 23-25, 27, 32, and 33 have been amended as suggested by the Examiner to more precisely define the claimed invention. These amendments derive support from throughout the specification and claims as originally filed. No new matter has been added.

Claim Rejections -35 35 U.S.C. § §112

Claims 1-25 and 27-33 are rejected under 35 U.S.C. § §112, second paragraph, as purportedly indefinite. At page 3 of the Official Action the Examiner asserts that the term "substance" renders the claims indefinite, and suggests that the term "protein" be substituted for "substance." Without conceding to the merits of this rejection, but solely in an effort to expedite prosecution, the claims have been amended as suggested by the Examiner. Withdrawal of this rejection is thus respectfully requested.

Claim Rejections - 35 U.S.C. § §102/103

Claims 1-4, 19-21, and 28-32 are rejected under 35 USC §102(b) as purportedly anticipated by, or in the alternative, under 35 U.S.C. § §103(a) as purportedly obvious over Wagner et al. (WO 93/02216). This rejection, to the extent that it applies to the claims as amended, is respectfully traversed.

In order to anticipate a claim under 35 U.S.C. §102b), a reference must teach every element of the claim. *See* MPEP 2131 *et seq.* The Examiner notes, at page 4 of the Official Action, that Wagner does not teach every element of the presently claimed invention, but argues that the missing feature - a full-length gene - is inherent in Wagner et al.

> Wagner is silent with regard to the fragment having all of a full-length gene. However, the sequence of a full-length gene recited in Claim 1 is deemed to be inherent in the DNA hybridization partner having a mRNA target in Wagner et al. because DNA hybridization partners of mRNA inherently encompass a full-length gene and therefore the DNA hybridization partners of Wagner et al. encompass the sequence of a full-length gene.

Applicant respectfully disagrees. Wagner et al teach, page 6, lines 26-27, that the hybridization partner is cDNA or a synthetic oligonucleotide. Then, at page 6, lines 27-28, that the hybridization target is mRNA. Accordingly, Applicant submits that in the paragraph (page 6, lines 25-28) Wagner exhibits the explicit intention to distinguish clearly between "partner" and "target". The hybridization partner in Wagner is a cDNA or oligonucleotide fragment and NOT a full-length gene. Applicant respectfully maintains that a full-length gene is a very specific case of cDNA and without any precise and specific reference to it, one of ordinary skill in the art would understand the reference to cDNA in Wagner et al. to refer to EST sequences or shotgun fragments and NOT to full-length genes.

This is confirmed in Wagner et al, page 44, Example III, and page 46, Example IV, where the preparation of the cDNA molecule used as hybridization partner (not target) are prepared by standards methods (Sambrook et al). Standard methods and Sambrook et al do NOT include the use of full-length genes as hybridization partners.

There is no reference, indication nor suggestion in Wagner et al to prepare full-length gene as a hybridization partner. On the contrary, the reference to standard method is a clear indication to explicitly exclude a full-length cDNA as hybridization target. Accordingly, because Wagner et al. does not disclose all of the limitations of the presently claimed invention, as required by 35 U.S.C. § §102(b), the present claims are not anticipated by Wagner et al.

The requirements of a *prima facie* case of obviousness are set forth in MPEP 2143:

> To establish a *prima facie* case of obviousness, three basic criteria must be met. First, there must be some suggestion or motivation, either in the references themselves or in the knowledge generally available to one of ordinary skill in the art, to modify the reference or to combine reference teachings. Second, there must be a reasonable expectation of success. Finally, the prior art reference (or references when combined) must teach or suggest all the claim limitations.

As discussed above, Wagner et al. neither disclose nor suggest the use of a DNA with the sequence of a full-length gene as a hybridization partner. The fact that Wagner et al neither disclose nor suggest a full-length sequence as hybridization partner may also be concluded because Wagner et al use the "tiling methodology" according to which several hybridization partner fragments are fixed on a support in order to correspond (as a group) to the complete sequence of a full-length gene. The availability of short fragments as partners makes it

possible to define the position of the mutated base (according to which "tile" binds the mutated position). The use of a full-length gene as hybridization partner is fundamentally incompatible with the "tiling methodology" carried out by Wagner. The presence of a full-length gene as hybridization partner is thus completely inconsistent with the use of fragments as hybridization partners. The "tiling methodology" and the "full-length" partner methodology are based on a different system and give different results.

The Examiner's attention is respectfully directed to the attached Declaration of Okazaki Yasushi, confirming these conclusions. Dr. Okazaki's declaration is being submitted in unsigned form; a signed copy of his declaration will be provided as soon as it is received by the undersigned.

Accordingly, for the foregoing reasons, Applicant maintains that the presently claimed invention is neither anticipated by, nor obvious over, Wagner et al. Withdrawal of this rejection is thus respectfully requested.

Claim Rejections - 35 U.S.C. §103

Claim 5 is rejected under 35 USC §103(a) as purportedly obvious over Wagner et al. (WO 93/02216) in view of Zoltukhin et al. (U.S. 5,874,304). This rejection, to the extent that it applies to the claims as amended, is respectfully traversed.

The requirements of a *prima facie* case of obviousness, and the deficiencies of Wagner et al., are set forth in detail above. The Examiner cites Zoltukhin et al. for its teaching of GFP labeled proteins (Official Action at 7). However, Zoltukhin et al. does not remedy the deficiencies of Wagner et al; specifically, Zoltukhin et al. neither discloses nor suggests the use of hybridization partners comprising the sequence of a full-length gene. Consequently, the presently claimed invention is not *prima facie* obvious over the combination of Wagner et al. and Zoltukhin et al., because these publications neither disclose nor suggest all of the limitations of the presently claimed invention, as required by 35 U.S.C. §103. Withdrawal of this rejection is thus respectfully requested.

Claims 6-8 are rejected under 35 USC §103(a) as purportedly obvious over Wagner et al. (WO 93/02216) in view of Gifford (U.S. 5,750,335). This rejection, to the extent that it applies to the claims as amended, is respectfully traversed.

The requirements of a *prima facie* case of obviousness, and the deficiencies of Wagner et al., are set forth in detail above. The Examiner cites Gifford for its teaching of a method

> for detecting nucleic acid fragment having a mutation comprising: hybridizing at least one fragment fixed on a substrate with at least on fragment of which mutation is to be assayed . . . and introducing a label into a fragment to be assayed to identify and quantify the fragment having a mismatch.

Official Action at 9. However, Gifford does not remedy the deficiencies of Wagner et al; specifically, Gifford neither discloses nor suggests the use of hybridization partners comprising the sequence of a full-length gene. Consequently, the presently claimed invention is not *prima facie* obvious over the combination of Wagner et al. and Gifford, because these publications neither disclose nor suggest all of the limitations of the presently claimed invention, as required by 35 U.S.C. § §103. Withdrawal of this rejection is thus respectfully requested.

Claims 9-18 and 33 are rejected under 35 U.S.C. § §103(a) as purportedly obvious over Wagner et al. (WO 93/02216) in view of Chirikjian et al. (U.S. 5,763,178) and Goldrick (U.S. 5,891,629). This rejection, to the extent that it applies to the claims as amended, is respectfully traversed.

The requirements of a *prima facie* case of obviousness, and the deficiencies of Wagner et al., are set forth in detail above. The Examiner cites Chirikjian et al. for its teaching of a method

> for detecting nucleic acid fragment having a mutation comprising: hybridizing nucleic acid fragments with nucleic acid fragments of which mutation is to be assayed treating a mismatched base pair occurring between the fragments with a substance specifically recognizing and cleaving the mismatched base pair labeling the cleaved fragments . . . and identifying the labeled fragment to thereby detect a nucleic acid having a mutation.

Official Action at 10. However, Chirikjian et al. does not remedy the deficiencies of Wagner et al; specifically, Chirikjian et al. neither discloses nor suggests the use of hybridization partners comprising the sequence of a full-length gene.

The Examiner cites Goldrick for its teaching of a method

> for detecting a mutation comprising: hybridizing a nucleic acid fragment with a fragment to be assayed; treating a mismatched base pair with a substance

specifically recognizing and cleaving the mismatch base pair to cleave; and identifying the cleaved fragment to identify the mutated fragment wherein the cleaving substance is selected from S1 nuclease and Mung bean nuclease.

Official Action at 11-12. However, Goldrick does not remedy the deficiencies of Wagner et al; specifically, Goldrick neither discloses nor suggests the use of hybridization partners comprising the sequence of a full-length gene.

Consequently, the presently claimed invention is not *prima facie* obvious over the combination of Wagner et al., Chirikjian et al., and Goldrick because these publications neither disclose nor suggest all of the limitations of the presently claimed invention, as required by 35 U.S.C. § §103. Withdrawal of this rejection is thus respectfully requested.

Claims 23-25 are rejected under 35 U.S.C. § §103(a) as purportedly obvious over Wagner et al. (WO 93/02216) in view of Zoltukhin et al. (U.S. 5,874,304) and Fleck et al. (*Nucl. Acids Res.*, 1994, 22(24):5289-5295). This rejection, to the extent that it applies to the claims as amended, is respectfully traversed.

The requirements of a *prima facie* case of obviousness, and the deficiencies of Wagner et al., are set forth in detail above. The Examiner cites Zoltukhin et al. for its teaching of GFP labeled proteins (Official Action at 14), and Fleck et al. for its teaching of "the MutS homologue of *Schizosaccharomyces pombe, swi4* which specifically binds to c/c mismatched base pairs" (Official Action at 14). However, neither Zoltukhin et al. nor Fleck et al. remedy the deficiencies of Wagner et al; specifically, neither publication discloses or suggests the use of hybridization partners comprising the sequence of a full-length gene. Consequently, the presently claimed invention is not *prima facie* obvious over the combination of Wagner et al., Zoltukhin et al., and Fleck et al. because these publications neither disclose nor suggest all of the limitations of the presently claimed invention, as required by 35 U.S.C. § §103. Withdrawal of this rejection is thus respectfully requested.

From the foregoing, further and favorable reconsideration in the form of a Notice of Allowance is believed to be next in order and such action is earnestly solicited.

In the event that there are any questions concerning this amendment, or the application in general, the Examiner is respectfully urged to telephone the undersigned so that prosecution of this application may be expedited.

Respectfully submitted,

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Attachment to Reply & Amendment dated June 25, 2001
Marked-up Claims 1, 2, 4, 5, 7-9, 11, 15, 18, 23-25, 27, 32, and 33

1. (Three times Amended) A method for detecting nucleic acid fragment and/or PNA having a mutation, comprising the steps of:
(A) hybridizing at least one fragment among one or more fragments fixed on a substrate, which fragments are selected from the group consisting of one or more nucleic acid fragments and one or more PNA fragments and have all of a sequence of full-length gene, with at least one fragment of which mutation is to be assayed, wherein said fragment is selected from the group consisting of one or more nucleic acid fragments and one or more PNA fragments;
(B) binding a labeled [substance] protein, said [substance] protein specifically binding to a mismatched base pair occurring between the hybridized fragments having a mutation; and
(C) identifying a fragment bound by the labeled [substance] protein by detecting the label, thereby detecting a nucleic acid and/or PNA fragments having a mutation..

2. (Amended) The method of claim 1, wherein the [substance] protein specifically binding to a mismatched base pair is a mismatch binding protein.

3. The method of claim 2, wherein the mismatch binding protein is Mut S protein or analogue thereof, or a C/C mismatch binding protein.

4. (Twice Amended) The method of claim 1, wherein the [substance] protein specifically binding to a mismatched base pair is labeled with at least one kind of [substance] protein selected from the group consisting of luminescent proteins, phosphorescent proteins, fluorescent proteins, [luminescent substances, fluorescent substances, phosphorescent substances,] radioactive [substances] proteins, stable isotopes, antibodies, antigens, and enzymes [and proteins].

5. (Twice Amended) The method of claim 1, wherein the [substance] protein specifically binding to a mismatched base pair is labeled with GFP (Green Fluorescence Protein).

6. (Twice Amended) The method of claim 1, wherein introducing a label into a nucleic acid and/or PNA fragment to be assayed for mutations, and detecting the label of the nucleic acid and/or PNA fragment to be assayed for mutations, are carried out in order to identify and quantify the fragment having a mismatched base pair.

7. (Amended) The method of claim 6, wherein the label introduced into the nucleic acid and/or PNA fragment to be assayed for mutations produce a signal different from that produced by the label attached to the [substance] protein specifically binding to the mismatched base pair, and quantification and identification of the fragment having a mismatched base pair are simultaneously performed.

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8. (Three times Amended) The method of claim 6, wherein the nucleic acid and/or PNA to be assayed for mutations is labeled with at least one kind of label selected from the group consisting of luminescent [substances] proteins, fluorescent [substances] proteins, phosphorescent [substances] proteins, stable isotopes, radioactive [substances] proteins, antibodies, antigens, and enzymes [and proteins].

9. (Twice Amended) A method for detecting a nucleic acid fragment and/or PNA fragment having a mutation, comprising the steps of:
(A) hybridizing at least one fragment among one or more fragments fixed on a substrate, which fragments are selected from the group consisting of one or more nucleic acid fragments and one or more PNA fragments and have all of a sequence of full-length gene, with at least one fragment of which mutation is to be assayed, wherein said fragment is selected from the group consisting of one or more nucleic acid fragments and one or more PNA fragments;
(D) treating a mismatched base pair occurring between the hybridized fragments with a [substance] protein specifically recognizing and cleaving the mismatched base pair to cut the hybridized fragments at the mismatched base pair, or to remove at least a part of one strand of the fragments hybridized from the mismatched base pair;
(E) labeling a fragment remained on the substrate after the cleavage or removal; and
(F) identifying the labeled fragment by detecting the label, thereby detecting a nucleic acid and/or PNA fragment having a mutation.

10. (Twice Amended) The method of claim 9, wherein said at least one fragment is fixed on the substrate at the 5' end and the 3' end of said fragment is blocked, and the labeling of the fragment in step (E) is performed by 3' end addition reaction.

11. (Twice Amended) The method of claim 9, wherein the [substance] protein specifically recognizing and cleaving the mismatched base pair is a nuclease.

12. The method of claim 11, wherein the nuclease is S1 nuclease, Mung bean nuclease or RNase H.

13. (Twice Amended) The method of claim 9, wherein the labeling of the fragment in the step (E) is performed by an enzyme reaction utilizing a label.

14. The method of claim 13, wherein the enzyme reaction is polymerase reaction, kination reaction, ligation reaction, or 3' end addition reaction.

15. (Three times Amended) The method of claim 13, wherein the fragment is labeled with at least one kind of label selected from the group consisting of luminescent [substances] proteins, fluorescent [substances] proteins, phosphorescent [substances] proteins, stable isotopes, radioactive [substances] proteins, antibodies, antigens, and enzymes [and proteins].

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16. (Twice Amended) The method of claim 9, wherein introducing a label into a nucleic acid and/or PNA fragment to be assayed for mutations, and detecting the label of the nucleic acid and/or PNA fragment to be assayed for mutations, are carried out in order to detect and quantify the fragment having a mismatched base pair.

17. The method of claim 16, wherein the label introduced into the nucleic acid and/or PNA fragment to be assayed for mutations produce a signal different from that produced by the label attached to the fragment in the step (E), and quantification and identification of the fragment having a mismatched base pair are simultaneously performed.

18. (Three times Amended) The method of claim 16, wherein the nucleic acid and/or PNA to be assayed for mutations is labeled with at least one kind of label selected from the group consisting of luminescent [substances] proteins, fluorescent [substances] proteins, phosphorescent [substances] proteins, stable isotopes, radioactive substances, antibodies, antigens, and enzymes [and proteins].

19. (Amended) The method of claim 1, wherein the fragments of nucleic acid or PNA fixed on the substrate are bound to the substrate only at their 5' or 3' end.

20. (Amended) The method of claim 1, wherein the fragments of nucleic acid or PNA fixed on the substrate are fixed on the substrate by covalent bonds.

21. (Twice Amended) The method of claim 1, wherein said nucleic acid or PNA is cDNA.

22. (Twice Amended) The method of claim 9, wherein said nucleic acid or PNA is cDNA.

23. (Twice Amended) A [substance] protein specifically bindable to a mismatched base pair wherein said [substance] protein is labeled with GFP (Green Fluorescence protein).

24. (Twice Amended) The [substance] protein of claim 23, wherein the [substance] protein specifically bindable to the a mismatched base pair is a C/C mismatch binding protein.

25. (Twice Amended) A [substance] protein specifically bindable to a mismatched base pair, wherein said [substance] protein is a C/C mismatch binding protein.

27 (Three times Amended) The [substance] protein of claim 25, wherein the label is at least one kind of label selected from the group consisting of luminescent proteins, phosphorescent proteins, fluorescent proteins, [luminescent substances, fluorescent substances, phosphorescent substances,] stable isotopes, radioactive [substances] proteins, antibodies, antigens, and enzymes [and proteins].

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28. (Twice Amended) An article comprising a substrate having a surface on which one or more kinds of nucleic acid or PNA fragments having all of the sequence of a full-length gene are fixed in a hybridizable condition.

29. (Amended) The article of claim 28, wherein said fragments fixed on the substrate are bound to the substrate only at their 5' or 3' ends.

30. (Twice Amended) The article of claim 28, wherein said fragments fixed on the substrate are bound to the substrate by covalent bonds.

31. The article of claim 28, wherein said nucleic acid or PNA is cDNA.

32. (Amended) A method for detecting nucleic acid and/or PNA having a mutation, comprising the steps of:
(A) providing
- at least one polynucleotide fixed on a substrate, wherein said polynucleotide has all of the sequence of full-length gene;
- a sample comprising at least one fragment of which mutation is to be assayed, wherein said fragment is selected from the group consisting of one or more nucleic acid fragments and one or more PNA fragments; and
- a labeled [substance] protein, wherein said [substance] protein specifically binds to a mismatched base pair resulting from hybridization between a polynucleotide and a fragment comprising a mutation;

(B) hybridizing said fragment to said polynucleotide;
(C) introducing said labeled [substance] protein under conditions that permit said [substance] protein to specifically bind to any mismatched base pairs that are present; and
(D) identifying a fragment bound by the labeled [substance] protein by detecting the label, thereby detecting a nucleic acid and/or PNA fragments having a mutation.

33. (Amended) A method for detecting a nucleic acid fragment and/or PNA fragment having a mutation, comprising the steps of:
(A) providing
- at least one polynucleotide fixed on a substrate, wherein said polynucleotide has all of the sequence of full-length gene; and
- a sample comprising at least one fragment of which mutation is to be assayed wherein said fragment is selected from the group consisting of one or more nucleic acid fragments and one or more PNA fragments;

(B) hybridizing said fragment to said polynucleotide;
(C) treating a mismatched base pair occurring between said hybridized fragment and said polynucleotide with a [substance] protein that specifically recognizes and cleaves a mismatched base pair to cut the hybridized nucleic acids at the mismatched base

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pair, or to remove at least a part of one strand of the nucleic acids hybridized from the mismatched base pair;

(D) labeling a polynucleotide remained on the substrate after the cleavage or removal; and

(F) identifying the labeled polynucleotide by detecting the label, thereby detecting a nucleic acid and/or PNA fragment having a mutation.

Curriculum vitae

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